ABSTRACT OF THE DISCLOSURE

The invention is directed to a variety of multiplexing methods used to amplify and/or genotype a variety of samples simultaneously. The invention provides a method of detecting a target sequence. The method consists of: (a) contacting a first and second probe with a target sequence under conditions where complementary probes form a hybridization complex with the target sequence, the first probe comprising an upstream universal priming site and a target-specific sequence, the second probe comprising a downstream universal priming site and a target-specific sequence, wherein one of the first or second probes comprise an adapter sequence; (b) extending the first or second probe of the hybridization complex to form a modified probe; (c) amplifying the modified probe to form an amplicon, and (d) detecting the amplicon. A method of detecting the relative amounts of two or more target sequences is also provided. The method consists of: (a) contacting a first and a second probe with first and second target sequences in an initial population under conditions where complementary probes form a hybridization complex with the target sequences, the first and second probes comprising a universal priming site, an adapter sequence and a target-specific sequence; (b) linearly amplifying the first and second probes forming the hybridization complex to produce first and second amplicons having distinctive adapter sequences, and (c) determining a relative amount of the first and second amplicons distinguishable by the adapter sequence, wherein the relative amount of the amplicons is indicative of the relative amounts of the first and second target sequences in the initial population. Further provided is a method of amplifying a target sequence to produce a signal within a dynamic range of a detection assay. The method consists of: (a) hybridizing a target-specific probe having an upstream universal priming site (UUP), a downstream universal priming site (DUP) and an adapter sequence with a set of differential primers, the set of differential primers comprising an upstream primer and first and second downstream primers, the second downstream primer having a lower Tm compared to the upstream primer and the first downstream primer; (b) amplifying the probe with the set of differential primers for two or more cycles of enzymatic polymerization; (c) increasing hybridization stringency to suppress hybridization of the second downstream primer, and (d) amplifying the probe from the upstream and the first downstream primers of the set for at least one cycle of enzymatic polymerization, wherein differential signals of amplicons produced from amplification of the first or the second downstream primers fall within a dynamic range of a detection assay.